

CLAIMS

What is claimed is:

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1. A method for identifying a test compound as a candidate for an antibiotic, comprising:

5 a) contacting a 3-Isopropylmalate dehydratase polypeptide with said test compound;
and *Isopropylmalate isomerase*
 hydro-lyase

 b) detecting the presence or absence of binding between a test compound and said 3-
Isopropylmalate dehydratase polypeptide,
wherein binding indicates that said test compound is a candidate for an antibiotic.

10 2. The method of claim 1, wherein said 3-Isopropylmalate dehydratase polypeptide is a
fungal 3-Isopropylmalate dehydratase polypeptide.

15 3. The method of claim 1, wherein said 3-Isopropylmalate dehydratase polypeptide is a
Magnaporthe 3-Isopropylmalate dehydratase polypeptide.

4. The method of claim 1, wherein said 3-Isopropylmalate dehydratase polypeptide is
SEQ ID NO: 3.

20 5. A method for determining whether the antibiotic candidate of claim 1 has antifungal
activity, further comprising:
contacting a fungus or fungal cells with said antibiotic candidate and detecting the
decrease in growth, viability, or pathogenicity of said fungus or fungal cells.

6. A method for identifying a test compound as a candidate for an antibiotic, comprising:

a) contacting a test compound with at least one polypeptide selected from the group consisting of: a polypeptide having at least ten consecutive amino acids of a fungal 3-Isopropylmalate dehydratase; a polypeptide having at least 50% sequence identity with a fungal 3-Isopropylmalate dehydratase; and a polypeptide having at least 10% of the activity of a fungal 3-Isopropylmalate dehydratase; and

5 b) detecting the presence and/or absence of binding between said test compound and said polypeptide,

wherein binding indicates that said test compound is a candidate for an antibiotic.

10 7. A method for determining whether the antibiotic candidate of claim 6 has antifungal

activity, further comprising:

contacting a fungus or fungal cells with said antibiotic candidate and detecting a decrease in growth, viability, or pathogenicity of said fungus or fungal cells.

15 8. A method for identifying a test compound as a candidate for an antibiotic, comprising:

a) contacting 2-Isopropylmalate and H₂O with a 3-Isopropylmalate dehydratase;

b) contacting 2-Isopropylmalate and H₂O with 3-Isopropylmalate dehydratase and a test compound; and

20 c) determining the change in concentration for at least one of the following: 2-

Isopropylmalate, H₂O, and/or 3-Isopropylmalate,

wherein a change in concentration for any of the above substances between steps (a) and

(b) indicates that said test compound is a candidate for an antibiotic.

9. The method of claim 8, wherein said 3-Isopropylmalate dehydratase is a fungal 3-
Isopropylmalate dehydratase.

5 10. The method of claim 8, wherein said 3-Isopropylmalate dehydratase is a
Magnaporthe 3-Isopropylmalate dehydratase.

10 11. The method of claim 8, wherein said 3-Isopropylmalate dehydratase is SEQ ID NO:
3.

12. A method for determining whether the antibiotic candidate of claim 8 has antifungal
activity, further comprising:
contacting a fungus or fungal cells with said antibiotic candidate and detecting a
decrease in growth, viability, or pathogenicity of said fungus or fungal cells.

15 13. A method for identifying a test compound as a candidate for an antibiotic,
comprising:

- a) contacting 3-Isopropylmalate with a 3-Isopropylmalate dehydratase;
- b) contacting 3-Isopropylmalate with a 3-Isopropylmalate dehydratase and a test
compound; and
- c) determining the change in concentration for at least one of the following: 2-
Isopropylmalate, H₂O, and/or 3-Isopropylmalate,

wherein a change in concentration for any of the above substances between steps (a) and (b) indicates that said test compound is a candidate for an antibiotic.

14. The method of claim 13, wherein said 3-Isopropylmalate dehydratase is a fungal 3-
5 Isopropylmalate dehydratase.

15. The method of claim 13, wherein said 3-Isopropylmalate dehydratase is a
Magnaporthe 3-Isopropylmalate dehydratase.

10 16. The method of claim 13, wherein said 3-Isopropylmalate dehydratase is SEQ ID NO:
3.

17. A method for determining whether the antibiotic candidate of claim 13 has antifungal
activity, further comprising:

15 contacting a fungus or fungal cells with said antibiotic candidate and detecting a
decrease in growth, viability, or pathogenicity of said fungus or fungal cells.

18. A method for identifying a test compound as a candidate for an antibiotic,
comprising:

20 a) contacting 2-Isopropylmalate and H₂O with a polypeptide selected from the group
consisting of: a polypeptide having at least 50% sequence identity with 3-
Isopropylmalate dehydratase; a polypeptide having at least 50% sequence identity
with a 3-Isopropylmalate dehydratase and having at least 10% of the activity

thereof; and a polypeptide comprising at least 100 consecutive amino acids of a 3-Isopropylmalate dehydratase;

b) contacting 2-Isopropylmalate and H₂O with said polypeptide and a test compound;

and

5 c) determining the change in concentration for at least one of the following: 2-

Isopropylmalate, H₂O, and/or 3-Isopropylmalate,

wherein a change in concentration for any of the above substances between steps (a) and

(b) indicates that said test compound is a candidate for an antibiotic.

10 19. A method for identifying a test compound as a candidate for an antibiotic, comprising:

a) contacting 3-Isopropylmalate with a polypeptide selected from the group consisting of: a polypeptide having at least 50% sequence identity with a 3-Isopropylmalate dehydratase; a polypeptide having at least 50% sequence identity with a 3-Isopropylmalate dehydratase and at least 10% of the activity thereof; and a polypeptide comprising at least 100 consecutive amino acids of a 3-Isopropylmalate dehydratase;

b) contacting 3-Isopropylmalate, with said polypeptide and a test compound; and

c) determining the change in concentration for at least one of the following: 2-

15 Isopropylmalate, H₂O, and/or 3-Isopropylmalate,

wherein a change in concentration for any of the above substances between steps (a) and

(b) indicates that said test compound is a candidate for an antibiotic.

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20. A method for identifying a test compound as a candidate for an antibiotic,
comprising:

5 a) measuring the expression of a 3-Isopropylmalate dehydratase in a cell, cells, tissue,
 or an organism in the absence of a test compound;

 b) contacting said cell, cells, tissue, or organism with said test compound and
 measuring the expression of said 3-Isopropylmalate dehydratase in said cell, cells,
 tissue, or organism; and

 c) comparing the expression of 3-Isopropylmalate dehydratase in steps (a) and (b),
wherein a lower expression in the presence of said test compound indicates that said test
compound is a candidate for an antibiotic.

15 21. The method of claim 20 wherein said cell, cells, tissue, or organism is, or is derived
 from a fungus.

20 22. The method of claim 20 wherein said cell, cells, tissue, or organism is, or is derived
 from a *Magnaporthe* fungus or fungal cell.

23. The method of claim 20, wherein said 3-Isopropylmalate dehydratase is SEQ ID NO:
 3.

24. The method of claim 20, wherein the expression of 3-Isopropylmalate dehydratase is
 measured by detecting IPMD1 mRNA.

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25. The method of claim 20, wherein the expression of 3-Isopropylmalate dehydratase is measured by detecting 3-Isopropylmalate dehydratase polypeptide.

26. A method for identifying a test compound as a candidate for an antibiotic, comprising:

5 a) providing cells having one form of a 3-Isopropylmalate dehydratase gene, and providing comparison cells having a different form of a 3-Isopropylmalate dehydratase gene; and

b) contacting said cells and said comparison cells with a test compound and determining the growth of said cells and comparison cells in the presence of the test compound,

10 wherein a difference in growth between said cells and said comparison cells in the presence of said compound indicates that said compound is a candidate for an antibiotic.

15 27. The method of claim 26 wherein the cells and the comparison cells are fungal cells.

28. The method of claim 26 wherein the cells and the comparison cells are *Magnaporthe* cells.

29. The method of claim 26 wherein said form and said comparison form of the 3-
20 Isopropylmalate dehydratase are fungal 3-Isopropylmalate dehydratases.

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30. The method of claim 26, wherein at least one of the forms is a *Magnaporthe* 3-
Isopropylmalate dehydratase.

31. The method of claim 26 wherein said form and said comparison form of the 3-
5 Isopropylmalate dehydratase are non-fungal 3-Isopropylmalate dehydratases.

32. The method of claim 26 wherein one form of the 3-Isopropylmalate dehydratase is a
fungal 3-Isopropylmalate dehydratase, and the other form is a non-fungal 3-
Isopropylmalate dehydratase.

33. A method for identifying a test compound as a candidate for an antibiotic,
comprising:
a) providing cells having one form of a gene in the L-leucine biochemical and/or
genetic pathway and providing comparison cells having a different form of said gene.
b) contacting said cells and said comparison cells with a said test compound,
c) determining the growth of said cells and said comparison cells in the presence of
said test compound,

wherein a difference in growth between said cells and said comparison cells in the
presence of said test compound indicates that said test compound is a candidate for
an antibiotic.

15 34. The method of claim 33 wherein the cells and the comparison cells are fungal cells.

35. The method of claim 33 wherein the cells and the comparison cells are *Magnaporthe* cells.

36. The method of claim 33 wherein said form and said different form of the L-leucine biosynthesis gene are fungal L-leucine biosynthesis genes.

37. The method of claim 33, wherein at least one form is a *Magnaporthe* L-leucine biosynthesis gene.

10 38. The method of claim 33 wherein said form and said different form of the L-leucine biosynthesis genes are non-fungal L-leucine biosynthesis genes.

15 39. The method of claim 33 wherein one form of the L-leucine biosynthesis gene is a fungal L-leucine biosynthesis gene, and the different form is a non-fungal L-leucine biosynthesis gene.

40. A method for determining whether the antibiotic candidate of claim 33 has antifungal activity, further comprising:

20 contacting a fungus or fungal cells with said antibiotic candidate and detecting a decrease in growth, viability, or pathogenicity of said fungus or fungal cells, wherein a decrease in growth, viability, or pathogenicity of said fungus or fungal cells indicates that the antibiotic candidate has antifungal activity.

41. A method for identifying a test compound as a candidate for an antibiotic,
comprising:

(a) providing paired growth media; comprising a first medium and a second medium,
wherein said second medium contains a higher level of L-leucine than said first
5 medium;

(b) contacting an organism with a test compound;

(c) inoculating said first and said second media with said organism; and

(d) determining the growth of said organism,

wherein a difference in growth of the organism between said first and said second media
10 indicates that said test compound is a candidate for an antibiotic.

42. The method of claim 41, wherein said organism is a fungus.

43. The method of claim 41, wherein said organism is *Magnaporthe*.

44. An isolated nucleic acid comprising a nucleotide sequence that encodes a
15 polypeptide of SEQ ID NO: 3.

45. The nucleic acid of claim 44 comprising the nucleotide sequence of SEQ ID NO: 1.

46. An expression cassette comprising the nucleic acid of claim 45.

47. The isolated nucleic acid of claim 44 comprising a nucleotide sequence with at least
50 to at least 95% sequence identity to SEQ ID NO: 1.

20 48. A polypeptide consisting essentially of the amino acid sequence of SEQ ID NO: 3.

49. A polypeptide comprising the amino acid sequence of SEQ ID NO: 3.